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Impact of germination versus non-germination on nutritional and functional potentials of two rice varieties (Ranjit and Ahu Kalogoria) available in three different districts of Assam, India

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ABSTRACT

Article History

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Keywords Ahu Kalogoria rice Germination Husk Nutritional value Ranjit rice Total flavonoid content Total phenolic content. The main purpose of this study is to explore the effect of germination and nongermination on the nutritional and functional properties of rice varieties. In this study, two types of rice-Ahu Kalogoria (germinated) and Ranjit (non-germinated)-and the husk of Ahu variety rice were chosen and described based on their accession numbers. The rice came from Goalpara, Chirang, and Kokrajhar in the Indian state of Assam. Finally, their calorific values and nutritional and functional constituents were examined. Standard and validated experimental protocols are considered for the assessment of all parameters during this study. RC-0648663 (Ranjit rice, Chirang) and LK1-0646984 (Ahu Kalogoria, Goalpara) contain higher calorific values, i.e., 361.24±1.95ªKcal/100gm and 356.35±1.32ªKcal/100gm, and the Husk LK1-0646984H (Ahu Kalogoria husk, Goalpara) contains 222.51±3.07 Kcal/100gm. Mg was found as a potential mineral in all varieties. Optimum total flavonoid content (TFC) and total phenolic content (TPC) were found in LK1-0646984(324.75±0.58^amg QE/100gm; mgGAE/100gm), LK1-0646984H $(345.82 \pm 1.57^{b} mgOE/100 gm;$ 353.7 ± 1.66^{a} 440.12±0.88b mg GAE/100gm) and RC-0648663 (69.375±1.3cmg QE (Quercetin equivalent) /100 gm; 106.79±0.54^c mgGAE/100gm) respectively. The DPPH (2, 2diphenyl 1-picrylhydrazyl) radical scavenging activity of LK1-0646984, LK1-0646984H, and RC-0648663 was determined to be maximum at 63.25±0.43a%, 69.36±0.31^b%, and 34.2±0.2^c%. Experimental results revealed that vitamin B5 content was higher in LK1-0646984 of 6.52mg/kg in comparison to RC-0648663 (Ranjit rice, Chirang) of 0.96mg/kg. Vitamin B1 was found at its maximum (3.53mg/kg) in LK1-0646984H. Therefore, the germinated rice variety (LK1-0646984) was found to be superior in quality to the non-germinated variety based upon nutritional, functional, and energy assessments, provide significant health benefits, and gain importance in the present socioeconomic scenario.

Contribution/Originality: Germination of rice provides health benefits by significantly improving the nutrients, viz., B vitamins and Mg content, and functional constituents, viz., TPC and TFC, and enhancing calorific value. Therefore, it can be considered an efficient and novel processing technique for the purpose of human consumption in comparison to its non-germinated counterpart.

1. INTRODUCTION

The two most popular cereals consumed worldwide are rice and wheat. It is the grain of certain kinds of grass species from the Oryza and Zizania genera (Carcea, 2021). If we look at the average paddy rice output by region from 1994 to 2020, we can see that Asia produced 90.5% of the total, followed by the Americas (5.2%), Europe (0.6%), Oceania (0.1%), and Africa (3.6%). Among the Asian regions, India is the second most producing region,

with about 143,740,297.26 tons. One of the most significant and consistent crops for human consumption is rice (Oryza sativa). Mainly in underdeveloped nations, rice is a major source of carbohydrates, protein, and other critical elements for billions of people globally (Huang et al., 2016; Parengam et al., 2010). After being harvested, rice grain is frequently put through a number of processing procedures, including drying, milling, and packaging to make it convenient for eating. The hull is taken out in the initial milling phase, and the whole brown rice grain (BR), which includes the outermost bran layer with a typically brown appearance, is obtained from the complete rice grain or paddy. To achieve polished or white rice (WR), the second stage involves removing the outer bran layer. Some of the layers that make up the bran are the pericarp, aleurone, sub-aleurone layer, and germ. These layers are full of nutrients and bioactive substances (Kaur, Ranawana, & Henry, 2016; Sharif, Butt, Anjum, & Khan, 2014). Protein, fat, vitamins, and minerals were often detected in greater quantities in Brown rice relative to White rice. While red, purple, and black-colored rice varieties are also known, white rice is still very common in South East Asia. In addition to genotypic variations, the grain micronutrient concentration is also dependent on location (Rao et al., 2014). Rice's nutritional value differs based on a number of factors, including the strain or variety (e.g., white, brown, red, and black/purple), the soil's nutritional status, the amount of milling, and the preparation method used before consumption, according to a detailed analysis of the nutritional value of rice (Devi, Padmavathi, Babu, & Waghray, 2015).

The period of storage of foods is significantly influenced by the moisture content (Ebuehi & Oyewole, 2008). A higher concentration of bioactive substances, including phenolic acids, flavonoids, α -tocopherol, and γ -tocotrienol, has been found in whole Brown rice grains (Saleh, Wang, Wang, Yang, & Xiao, 2019). The nutritive value of rice varies substantially based on both variety and environmental circumstances. Even a similar variety of rice may display nutritional variations due to different soil and climatic conditions. Brown rice's main ingredients are 0.8-2.6% fiber, 1.5-2.1% ash, 2.4-3.9% fat, 15.4% protein (N 5.95), and 1520-1610 kJ/100 g of calories (Bhattacharya, 2017; Zheng & Lan, 2007). The moisture content of different rice types depends on their genetic composition and the environmental conditions under which they are grown (Rathna Priya, Eliazer Nelson, Ravichandran, & Antony, 2019; Zheng & Lan, 2007). Brown rice can be enhanced in quality using a number of techniques, including germination, milling, enzymatic treatment, and pre-soaking (Carcea, 2021). Protein, which is rice's second-most important ingredient after starch, affects both its flavor and nutritional value (Kennedy & Burlingame, 2003). Due to its higher lysine concentration than wheat, corn, millet, and sorghum, rice has a comprehensive amino acid profile, making its protein preferable to that of other cereal grains (Eggum, 1979). The rice bran (20%, dry basis) is where the majority of the lipids or fats are found in rice. Dietary fiber boosts the volume of faeces, which has a laxative impact on the digestive system. Well-milled rice contains 0.5-1.0% fiber (Oko & Onyekwere, 2010). The rice variety, level of the milling process, and water solubility all affect the level and percentage of non-starch polysaccharide (Lai, Lu, He, & Chen, 2007). Variable rice cultivars may have variable levels of ash due to genetic or soil mineral composition considerations. Rice contains calcium, magnesium, iron, and zinc, which are the most frequently occurring minerals. Magnesium contributes to the insulin-mediated regulation of glucose absorption and increases insulin sensitivity (Andarini, Kiwari, & Handayani, 2022). The type of rice and level of machining, which entirely or partially eliminate the bran layer, aleurone layer, and embryo, affect the proximate composition of rice and its components. As a result, the nutritional value of the different rice portions from the same rice variety varies. The changes can be seen in the grain's content of minerals, fiber, and fats.

Since rice is consumed in a wide range of ways, multiple rice delicacies have been made utilizing unique approaches. There are several procedures that are used effectively on the grain, such as parboiling, annealing, and fast cooking methods, as well as some that encourage germination (Do Nascimento, Abhilasha, Singh, Elias, & Colussi, 2022). The popular and efficient process of germination enhances the nutritive value of cereal grains. External variables that influence the germination process include the germination period and the availability or lack of light, both of which can promote or prevent germination depending on the reserve (nutrient content) in the seed

(Lee et al., 2019). It has been determined that germination is a low-cost and useful technique for enhancing cereal nutrition. It has been used for generations to alter the structure of grain seeds' kernels, boost their nutrient content, and reduce their antinutritional content (Wu, Yang, Touré, Jin, & Xu, 2013). Based on farming methods, the growing season, and environmental considerations, paddy in Assam can be commonly separated into four groups. Example: Autumn rice (Ahu, from February to April/early June. Winter rice, also known as kharif (Sali: July to November/December), is harvested in the spring; Boro is harvested between November and May; and Bao is harvested from March to November/December. Of them, kharif rice, also known as sali, dominates with 66% of the paddy acreage and 73% of the rice yield (Dasgupta & Handique, 2018). The point of this study was to look at the nutritional value and usefulness of two types of rice from Assam, India: Ahu Kalogoria (germinated) and Ranjit (non-germinated). The rice came from three different districts: Kokrajhar, Goalpara, and Chirang. The husk was also looked at; it is made up of germinated roots and shoots. The goal was to choose a good variety for further research.

2. MATERIALS AND METHOD

2.1. Collection of Rice

From the local farmers in Goalpara, Chirang, and Kokrajhar districts of Assam, India, two varieties of rice paddy (Oryza sativa) were randomly selected, called Ranjit rice and Ahu Kalogoria rice, and then handed over to Delhi ICAR (Indian Council of Agricultural Research) for the accession numbers. The Ranjit varieties are LK2-0646984 (from Goalpara), RC-0648663 (from Chirang), and RK-0648664 (from Kokrajhar). The Ahu Kalogoria Varieties are LK1-0646984 (from Goalpara), AC-0648665 (from Chirang), and AK-0648666 (from Kokrajhar).

2.2. Germination of Ahu Kalogoria Paddy Rice Grains

Whole rice grains were cleaned, steeped in distilled water (1:2) for 12 hours at 26–30 °C (room temperature), and then kept on a piece of wet cloth stored in the dark. Then cover it with the jute bag and place the banana leaves over it to keep it moistened. During the germination period, to maintain the grains in a humid environment, water was sprayed on them. After 5 days of germination, the roots and shoots became 2–5 cm and 0.5–1.5 cm in length. Then, to stop germination, grains were solar dried (Veluppillai, Nithyanantharajah, Vasantharuba, Balakumar, & Arasaratnam, 2009).

2.3. Preparation of Flour

Ahu grains that have been germinated and processed in Dheki (husking peddle) are used to thresh rice grains to remove the outer layer and break the roots and shoots. The rice was then separated from the mixture using a winnowing fan called a Kula in Assamese, which is constructed of bamboo. All the varieties of rice powdered into flour and the mixture of husks were also kept for further analysis.

2.4. Proximate Analysis

2.4.1. Determination of Crude Protein

The kjeldahl method was used to find the total nitrogen, which was then used to estimate the crude protein. The protein percentage was found by multiplying the N% by 5.95 (Horwitz, 2000).

2.4.2. Determination of Lipids

The sample was extracted with petroleum ether in a soxhlet apparatus for eight hours to get an idea of its lipid content. The amount of lipid was then measured after the petroleum ether was taken away (Horwitz, 2000).

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2.4.3. Determination of Ash

Ash content was determined by ashing the sample at 5500c for 6 hours in Muffle furnace (Horwitz, 2000). Ash (%) = weight of Ash – the weight of the sample $\times 100$

2.4.4. Determination of Crude Fiber

Crude fiber was estimated by the following method: At first, a 2 g sample was dissolved for 30 minutes in 0.255 N sulfuric acid. Following filtering, what remained was suspended in 0.313 N sodium hydroxide and heated for an additional 30 minutes. Thereafter, it was washed with 1.25% boiling sulfuric acid, three 50-mL volumes of distilled water, and 25 ml of ethanol. The residue was then placed in a crucible that had already been pre-weighed, dried at 130 °C, weighed after cooling, and fired for 30 minutes at 600 °C. After cooling, the crucible's final weight was measured (Saikia, Dutta, Saikia, & Mahanta, 2012). The crude fiber was estimated as

Crude fiber % = weight of crucible with dry residues – weight of crucible with ash/weight of sample \times 100

2.4.5. Carbohydrates Content

Available carbohydrate was obtained by difference (Saikia et al., 2012)-

Carbohydrate (%) = 100 - (protein + Ash + Moisture + fat + Fiber)

2.4.6. Determination of Total Calorific Value

The calorific value of meals is a crucial characteristic that reveals their usable energy content. Osborne and Voogt (1978) computed the gross food energy using the equation:

Food energy $(kCal/g) = (CP \times 4) + (F \times 9) + (CHO \times 4)$

Where CP means crude protein (%); F means fat (%); and CHO means carbohydrate content (%).

2.4.7. Estimation of Minerals

The mineral analysis of the rice sample was determined by using atomic absorption spectrometry (AAS). In this determination, acid digestion of the sample is required, and the digestion process was carried out by nitric acid a (HNO_3) and chloric acid $(HClO_4)$ mixture in a 2:1 ratio (Uddin et al., 2016). 5 ml of 65% HNO_3 was added to the sample, and the mixture was boiled gently for 30–45 min. 2.5 ml of 70% $HClO_4$ was added after cooling, and the mixture was gently boiled until dense white fumes appeared. Now, the mixture was allowed to cool, and 10 ml of deionized water was added, followed by further boiling until the fumes were totally released. During the digestion procedures, the inner walls of the beakers were washed with 2 ml of deionized water to prevent the loss of the sample, and at the end of the digestion processes, the samples were filtered with Whatman 42 (2.5-µm particle retention) filter paper. Then, a sufficient amount of deionized water was added to make the final volume up to 50 ml. Finally, the digested sample was analyzed for minerals.

2.4.8. Determination of Total Phenolic Content (TPC)

A modified colorimetric technique was used to determine the total phenolic content (Ti et al., 2014). In this method, 120μ l each of extract and 2.5 ml of Folin-Ciocalteu reagent (10%) were mixed well, and within 8 min, 2 ml of 7.5% sodium carbonate (Na₂CO₃) was added. The samples were vortexed immediately and allowed to incubate in the dark for 30 minutes at 40 °C. Using a Shimadzu UV-1800 spectrometer (Shimadzu Inc., Kyoto, Japan), the absorbance of the ensuing blue colour was measured at 760 nm. The total amount of phenolics was given as mg of gallic acid equivalents (GAE) per 100 g of sample dry weight, with gallic acid (GA) serving as the standard dry weight (DW).

2.4.9. Determination of Flavonoid Content (TFC)

A modified technique (Dewanto etal. 2002) was used to determine the total flavonoid content (TFC). A stock solution of the crude extract of the sample is prepared by using methanol (1 mg/ml). 200 microliters of sample were taken from the stock solution. Make up the volume up to 1ml in all the test tubes with methanol, followed by the addition of 0.5 ml of 5% NaNO₂ and 0.5 ml of 10% AlCl₃. After 5 minutes of reaction, 2 ml of NaOH (4%) solution was added and incubated at room temperature for 15 minutes. The absorbance was observed at 510nm using a UV-VIS (Ultra Violet Visible) spectrophotometer. Quercetin is used as a standard solution. Blank is prepared by using methanol (1 ml) and all chemicals except the sample solution. The total flavonoid content (TFC) is expressed as mg quercetin equivalent (QE)/100 g.

2.4.10. Determination of Antioxidant Activity by DPPH Method

This method was described by Dewanto, Wu, Adom, and Liu (2002), with some modifications. DPPH gives strong absorbance at 517nm (deep violet colour) due to its unpaired electron. When this radical pairs off in the presence of a free radical scavenger, the absorption vanishes, resulting in decolouration or a yellowish colour. 200-microliter sample extract and make up the volume up to 1 ml with methanol, and control 1 ml methanol and 3 ml DPPH (0.004%).

DPPH scavenged% = Acon-Atest / Acon \times 100

Acon - is the absorbance of the control reaction.

Atest – is the absorbance of the test sample.

2.5. Sample Preparation for the Vitamin B complex Analysis

Put accurately weighed 5 g of ground powder into 25 mL of water. After 45 minutes of ultrasonic extraction, the samples were centrifuged at 4,500 rpm. Then the supernatant was filtered through a $0.4 \mu m$ filter, and the samples were injected into the HPLC system (Technical Note 7248, 2017).

2.5.1. HPLC Condition

The analysis of vitamin B complex in rice and husk was done using HPLC-DAD (High-Performance Liquid Chromatography with Diode-Array Detection) with column Shim-pack (4.6 mm \times 150 mm, 3µm). The mobile phase was 25 mm HK₂PO₄ (pH-7) (solvent A) and Acetonitrile: methanol 50:50 (solvent B), with a flow rate of 0.45 ml/min and an injection volume of 20µl. The column temperature was maintained at 35°C.

2.6. Standard and Sample Preparation for the Vitamin E Analysis

Prepare working standards from 5 to 100 ppm. Store the stock standard solutions at 4 °C when not in use; also, store the stock standards of fat-soluble vitamins in the dark.

Weigh 2.5g of ground powder from the samples, then add 10 ml of methanol and acetonitrile (1:1, v/v) to the mark. Keep the sample in a sonicator for 45 minutes. Prior to injection, filter the solutions through a 0.2 µm filter (Joseph, 2011; Wawan, 2010).

2.6.1. HPLC Condition

The analysis of vitamin E in rice and husk was performed using an HPLC with column Shim-pack XR-ODS (Extended Reality Octadecyl-Silica)/C8/Phenyl (7.5 mm \times 3 mm, 2.2 µm). The mobile phase was 90% methanol (solvent A) and 10% water (solvent B), isocratic elution, with a flow rate of 0.7 ml/min and an injection volume of 20 µl. The eluate was detected using a DAD detector with a wavelength of 273 nm. The column temperature was maintained at 33°C.

3. STATISTICAL ANALYSIS

The experimental data were reported as the mean values and standard deviations. Mean comparisons among the treatments were performed using the Tukey's test at a significance level of p < 0.05. The analyses were performed using OriginPro 9.0 software and were performed in triplicate.

4. RESULT AND DISCUSSION

4.1. Proximate Analysis

4.1.1. Moisture Content

Moisture content is the key factor determining the shelf life of foods. It was seen that the range of the moisture content for Ranjit rice was from $9.92\pm0.06^{\rm bc}$ to $10.61\pm0.005^{\rm a}$ % and in Ahu Kalogoria rice was $8.52\pm0.01^{\rm e}$ to $10.4\pm0.21^{\rm ab}$ % (Table 1). RK-0648664 has the highest moisture content $10.61\pm0.005^{\rm a}$ % while LK1-0646984 contains $(8.52\pm0.01^{\rm e}$ %) lowest in all rice. The husk with the highest moisture content (9.780.06 bc %) is LK1-0646984H. This is less than the permissible moisture level (14%) for the safe storage of processed rice (Verma & Srivastav, 2017). All varieties were discovered to have moisture contents that were practically within the permitted limit (12%) for rice storage over an extended period of time (Adair et al., 1973). The variation in moisture content in the rice accessions may be the reason for the variation in moisture content in the harvested paddy (Asaduzzaman et al., 2013). According to (Rathna Priya et al., 2019), both a rice cultivar's genetic make-up and the environment in which it grows have an impact on its moisture content. This study is similar to the results found by Oko and Onyekwere (2010), Maisont and Narkrugsa (2009), and Moongngarm and Saetung (2010).

4.1.2. Ash Content

From Table 1, the highest ash content was observed in Ahu Kalogoria rice (LK1-0646984) of $2.00\pm0.02^{\circ}$ %.However, the Ranjit rice variety (RK-0648664) was shown 0.82 ± 0.01^{d} %. This result is comparable with the findings of Saikia et al. (2012) and Chinma, Anuonye, Simon, Ohiare, and Danbaba (2015). The ash content of AC-0648665H was found to be 10.27 ± 0.13^{a} %. According to (Shayo, Mamiro, Nyaruhucha, & Mamboleo, 2006), variations in the mineral content of the soils and irrigation water may be to blame for the differences in ash composition between all rice varieties. According to Moongngarm and Saetung (2010), the treatment of the water source, the use of fertilizer, variety, processing, and management of storage are just a few of the variables that affect variation in chemical content among rice. Ash content serves a key function in indicating the mineral components of a food sample (Bhat & Sridhar, 2008; Mbatchou & Dawda, 2013) and provides guidance to figure out how much of the important minerals are present in the food (Edeogu, Ezeonu, Okaka, Ekuma, & EIom, 2007).

4.1.3. Fat Content

Among Ranjit rice, LK2-0646985 has the greatest fat level 1.82 ± 0.06^{bc} % and AK-0648666 has the highest, 2.49 ± 0.01^{a} % in the Ahu Kalogoria variety rice, and LK1-0646984H contains a high fat content $(1.23\pm0.11^{de}$ %) in (Table 1). The results of this study are consistent and somewhat similar to previous research by Chatterjee and Das (2019); Moongngarm and Saetung (2010); Oko and Ugwu (2011); and Chinma et al. (2015). The values found in the study by Heinemann, Fagundes, Pinto, Penteado, and Lanfer-Marquez (2005), which revealed values for crude fat brown rice that varied from 2.37% to 3.02%. Rice fat comprises no cholesterol but is an excellent source of linoleic and other necessary fatty acids (Eggum, 1979). Cooked rice's flavor is influenced by its fat content, considering rice with a significant amount of fat prefers to be more delicious and contain fewer starches (Taira, Taira, & Fujii, 1979). As the majority of the fat in rice accumulates in the aulerone layer of the kernel, variations in the degree of milling may be the cause of distinctions in the fat content of rice accessions (Wang et al., 2006). Given that the majority of the fat in rice grains is unsaturated, which readily oxidizes when exposed to atmospheric oxygen, variations in the fat content of rice accessions may be caused by the oxidation of fat (Verma & Srivastav, 2017).

4.1.4. Protein Content

In the current research, the rice varieties varied in terms of crude protein; RK-0648664contains the higher content 11.3±0.18^b% and AK-0648666 contained more proteins 14.32±0.17^a% (Table 1). In husk LK1-0646984H, the protein concentration ranges from 5.9±0.21°% to 6.8±0.38°%. The amount of protein in the present investigation was similar to that discovered by Chatterjee and Das (2019) and Dasgupta and Handique (2018). Kennedy and Burlingame (2003) found that the protein quantity in unpolished rice (brown rice) in O. sativa types ranged from 4.5% to 15.9%. This study's protein content was comparable to their findings. The range of protein variance was 8.70% to 11.18% recorded by Dasgupta and Handique (2018), 9.17 to 11.77% by Resurrection, Juliano, and Tanaka (1979). Protein content is regarded as a measure of the nutritional quality of rice, and a cultivar deemed to have a protein content of 10% or higher is considered a good source of protein (Devi et al., 2015). In north-eastern hill states, indigenous cultivars have a high protein level that ranges from 6.14 to 12.07% (Devi et al., 2015; Thongbam et al., 2010). Various rice accessions may have distinct protein contents due to a variety of factors, including availability of water, management, fertilizer usage (soil nitrogen availability), environmental stress (such as salinity and alkalinity, temperatures, and diseases), the site of cultivation areas, conditions during cultivation, and time, all of which tend to raise the protein content of grains (Burešová, Sedláčková, Faměra, & Lipavský, 2010). In between the varieties of rice, AK-0648666H includes a lot of protein. This might be because germination turns on a lot of enzymes and some nitrogen compounds that aren't proteins, like nucleic acids. This can lead to a rise in protein levels (Traoré, Mouquet, Icard-Vernière, Traore, & Trèche, 2004).

4.1.5. Crude Fiber Content

In the present study for Ranjit rice landraces, crude fiber content varied from 0.98 ± 0.02^{d} in RC-0648663 to 1.62 ± 0.13^{cd} % in LK2-0646985, while the corresponding range of variation for Ahu Kalogoria rice is 2.09 ± 0.07^{cd} % to 3.02 ± 0.20^{c} % in AC-0648665 and LK1-0646984. LK2-0646985 and LK1-0646984 contain a higher amount of crude fiber (Table 1). The crude fiber content of the Husk is range from 23.94 ± 0.30^{b} % to 26.01 ± 0.52^{a} %. This study is similar to the findings of Anjum, Pasha, Bugti, and Butt (2007); Chatterjee and Das (2019); Dasgupta and Handique (2018); and Ebuehi and Oyewole (2008). Rice that has been properly processed typically contains 0.5% to 1.0% fiber (Oko & Onyekwere, 2010). In diabetics, fiber has the power to lower blood sugar and cholesterol levels after eating. Fiber helps prevent constipation and lowers the chance of bowel diseases. The lack of or insufficient intake of fiber in food may be linked to the high prevalence of a number of illnesses in humans (Verma & Srivastav, 2017). Fiber increases the volume of faeces, which has a cathartic impact on the digestive system (Mbatchou & Dawda, 2013).

4.1.6. Carbohydrate Content

All the varieties have distinct carbohydrate contents, with RC-0648663 and LK1-0646984 having the higher quantities of carbohydrates, i.e., $75.98\pm0.49^{a}\%$ and $70.15\pm0.20^{b}\%$. In husk, AK-0648666H has the high carbohydrate content of $49.44\pm0.29^{c}\%$ (Table 1). This study's results are comparable to those of Abubakar, Yakasai, Zawawi, and Ismail (2018); Chatterjee and Das (2019); Dasgupta and Handique (2018); and Sompong, Siebenhandl-Ehn, Linsberger-Martin, and Berghofer (2011), but slightly lower than those (Moongngarm & Saetung, 2010; Saikia et al., 2012). Due to the bran layer's presence, which primarily includes non-starch ingredients, the amount of carbohydrates may vary. According to Mbatchou and Dawda (2013), high starch content causes the individual grains to stick to one another, while a low starch content effectively stops the grains from adhering to one another after cooking. The removal of the majority of the other nutrients along with the bran during milling may be the cause of the variation in total carbohydrate content, the aleurone layer, and the germ of the rice, leaving only carbohydrates as the grain's sole primary nutrient (Chaiyasut et al., 2017). A rise in amylase activity may have

contributed to the decrease in total carbohydrate that came about as a result of increased germination time (Moongngarm & Khomphiphatkul, 2011).

4.1.7. Food Energy

Food energy values were appreciably different among all the Ranjit and Ahu Kalogoria rice varieties from different districts. The high value of food energy in Table 1 is in RC-0648663, LK1-0646984, and AK-0648666H i.e. 361.24 ± 1.95^{a} %, 356.35 ± 1.32^{a} % and 233.88 ± 2.53^{b} %. These findings are almost similar to or within the range to the findings by Ebuehi and Oyewole (2008). Food energy is a measurement of the volume of energy that can be gained from food through cellular respiration (Thomas, Wan-Nadiah, & Bhat, 2013).

Table 1. Proximate composition of rice varieties (g/100 g DM basis).

Sample	Moisture	Ash	Fat	Protein	Crude fiber	Carbohydrate	Energy (Kcal
							per/100gm)
RK-0648664	10.61 ± 0.005^{a}	0.82±0.01 ^d	1.60 ± 0.16^{bcd}	11.3 ± 0.18^{b}	1.29 ± 0.08^{d}	$74.38 {\pm} 0.23^{a}$	357.15 ± 1.56^{a}
LK2-0646985	10.36 ± 0.27^{ab}	0.66 ± 0.03^{d}	1.82 ± 0.06^{bc}	10.6 ± 0.22^{b}	1.62 ± 0.13^{cd}	$74.94{\pm}0.17^{a}$	358.54 ± 2.16^{a}
RC-0648663	9.92 ± 0.06^{bc}	0.69 ± 0.02^{d}	1.52 ± 0.01^{cd}	10.91 ± 0.12^{b}	0.98 ± 0.02^{d}	$75.98 {\pm} 0.49^{a}$	361.24 ± 1.95^{a}
AK-0648666	10.27 ± 0.19^{ab}	$1.98 \pm 0.09^{\circ}$	2.49 ± 0.01^{a}	14.32 ± 0.17^{a}	2.14±0.09 ^{cd}	68.8 ± 0.35^{b}	354.89 ± 1.74^{a}
LK1-0646984	8.52±0.01 ^e	2 ± 0.02^{c}	2.10 ± 0.10^{ab}	14.2 ± 0.16^{a}	$3.02 \pm 0.20^{\circ}$	70.15 ± 0.20^{b}	356.35 ± 1.32^{a}
AC-0648665	10.4±0.21 ^{ab}	1.98±0.15°	2.33 ± 0.18^{a}	13.98 ± 0.23^{a}	2.09 ± 0.07 ^{cd}	69.21 ± 0.27^{b}	353.76 ± 3.12^{a}
AK-0648666H	9.43 ± 0.06^{cd}	9.4 ± 0.31^{b}	0.99 ± 0.02^{e}	6.8±0.38 ^c	23.94 ± 0.30^{b}	49.44±0.29°	$233.88 \pm 2.53^{\rm b}$
LK1-0646984H	9.78 ± 0.06^{bc}	10.12±0.13 ^a	1.23 ± 0.11^{de}	5.9±0.21°	26.01 ± 0.52^{a}	46.96 ± 0.55^{d}	222.51±3.07°
AC-0648665H	8.98 ± 0.03^{de}	10.27 ± 0.13^{a}	0.89 ± 0.07^{e}	6.4 ± 0.24^{c}	25.02 ± 0.53^{ab}	48.44±0.30 ^{cd}	227.37 ± 0.73^{bc}

Note: The values are expressed as means ± SD of triplicate assays, and the values with different superscript (a,b,c,d,e) indicates that they are significantly different (p<0.05).

(Here H indicates for Husk mixture).

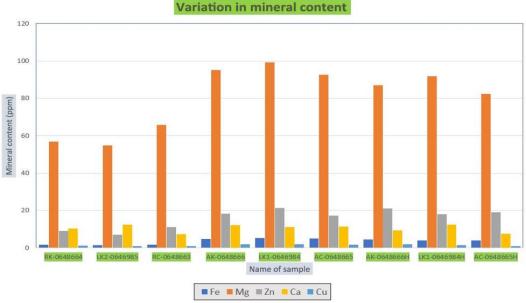


Figure 1. Variation in mineral content in variety of rice accessions in ppm.

4.2. Minerals

Mineral compositions were examined in all rice varieties, and the results are displayed in (Figure 1). Mg was to be found as a prevalent element in all rice varieties, with ranges of 99.3175 to 54.8851 ppm. All the rice varieties have various concentrations of Fe, Mg, Zn, and Ca in the range of 1.5212 to 5.3392, 54.8851 to 99.3175, 7.1178 to 21.2958, and 7.3854 to 12.3726 ppm, respectively. The most significant Mg concentrations are found in RC-0648663, LK1-0646984, and LK1-0646984H, with corresponding values of 65.906, 99.3175, and 91.8845 ppm. Abbas et al. (2011) found the result using the AAS method for Ca; the measured amount of Ca varies from 0.010 ppm to 0.090 ppm; the level of Fe varies from 0.030 ppm to 0.10 ppm; and for Zn, it varies from 0.040 ppm to 0.12

ppm which is lower than the present study. The mineral concentrations are either near or below the findings by Anjum et al. (2007), Verma and Srivastav (2017), and Sompong et al. (2011). The mineral composition of the soil or genetic factors may be to blame for these variations (Oko, Ubi, Efisue, & Dambaba, 2012). It has been demonstrated that the mineral element levels of rice are influenced by the quantity of fertilizer application and the inherent fertility of agricultural areas. Mineral content in rice is influenced by both genetic traits present in accessions and environmental variables (Zimmermann & Hurrell, 2002). However, Jiang, Wu, Feng, Yang, and Shi (2007) found that genotypic variations may offer chances to select for rice germplasm with higher mineral elements. Magnesium aids in the assimilation of calcium and potassium and contributes to the development of bones and teeth. Unlike calcium, which activates the muscles, magnesium makes the muscles relax. Through its assistance with enzyme activity, it is additionally required for metabolic processes in cells and the generation of energy (Mbatchou & Dawda, 2013). According to Chinma et al. (2015), the decrease in phytic acid concentration may be responsible for the greater mineral level of germinated rice flour in comparison with non-germinated samples. Phytic acid is a strong mineral inhibitor because it can strongly bind to cations, especially divalent cautions like Zn2+, Fe2+, Ca2+, and Mg2+, to make insoluble salts that stop the minerals from being bioavailable.

4.3. The Phenolic, Flavonoid Content, and DPPH-Scavenging Activity

LK1-0646984, LK1-0646984H, and RC-0648663 have respective total flavonoid contents of 324.75±0.58^a, 345.82±1.57^b, 69.375±1.3^c mgQE/100g, and phenolic contents of 353.7±1.66^a, 440.12±0.88^b, and 106.79±0.54^c mg GAE/100g. (Table 2) displays the DPPH scavenging activity at 63.25±0.43^a% in LK1-0646984, 69.36±0.31^b% in LK1-0646984H, and 34.2±0.2^c% in RC-0648663. These results are equivalent to those from Sompong et al. (2011), Abubakar et al. (2018), and Saikia et al. (2012), which are similar to or superior to the findings by Chinma et al. (2015), Ti et al. (2014), and Moongngarm and Saetung (2010). The level of TPC variation could be due to the presence or absence of the bran layer, and it was also affected by the main types of phenolic compounds found in the different types of rice (Moongngarm & Saetung, 2010). Different rice cultivars' varying total phenol content could be influenced by genetics, growing methods, and environmental factors present throughout the ripening process (Asaduzzaman et al., 2013). It has been suggested that the soaking and germination of cereals might generate enzymes that help break down the cell walls, enclosing numerous components. The release of free phenolic acids and bound phenolic acids may subsequently cause an increase in total phenolic acid levels (Kaukovirta-Norja, Wilhelmson, & Poutanen, 2008). The production of phenolic compounds triggered by enzyme hydrolysis during the germinating process may be the source of the greater total phenolic content of the germinated rice samples compared to that in non-germinated rice extracts (Lee et al., 2019).

Table 2. Total	phenolic conten	t, flavonoid content	and dpph scave	nging activity.

Sample name	Flavonoid content Mgqe/100g	Total phenolic content (Mggae/100g)	Dpph scavenging activity %
LK1-0646984	$324.75 {\pm} 0.58^{a}$	353.7 ± 1.66^{a}	$63.25 {\pm} 0.43^{a}$
LK1-0646984H	$345.82 \pm 1.57^{ m b}$	440.12 ± 0.88^{b}	69.36 ± 0.31^{b}
RC-0648663	69.375±1.3°	$106.79 \pm 0.54^{\circ}$	$34.2 \pm 0.2^{\circ}$

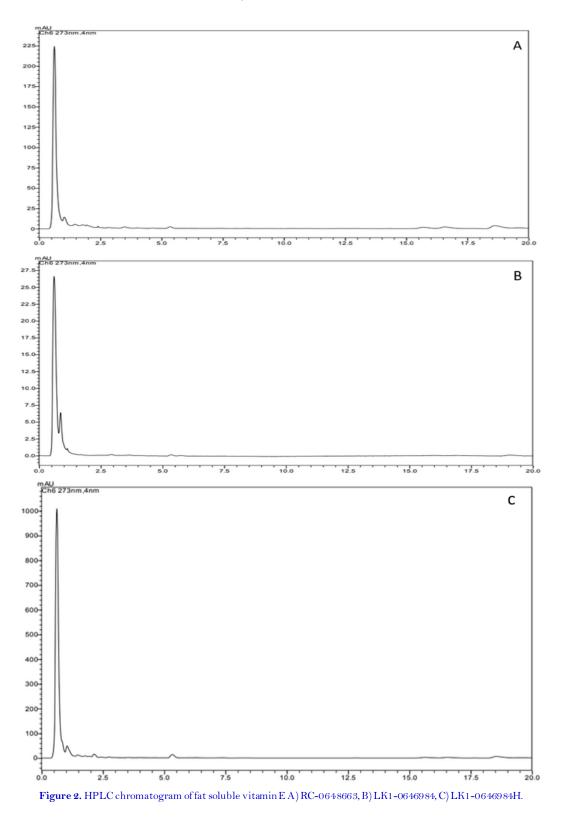
Note: The values are expressed as means \pm SD of triplicate assays, and the values with different superscripts (a,b,c) indicates that they are significantly different (p < 0.05).

4.4. Vitamin Content

Vitamin B5 is present in higher concentrations in RC-0648663 and LK1-0646984 (0.96 mg/kg and 6.52 mg/kg, represent in (Table 3 and 4) (Figure 3 and 4). B6, B9, and B2 levels in RC-0648663 are below detection level. Vitamins B1, B3, B12, and B9 are below detection level in LK1-0646984in (Table 4 and Figure 4). Vitamin B1 showing in (Table 5 and Figure 5) levels in LK1-0646984H are greater (3.53 mg/kg), but levels of B5, B6, B3, and B12 are below detection in (Table 5 and Figure 5). Vitamin E levels in all of the samples are below detection level

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described in (Figure 2). These findings are comparable to those of Prasad, Hymavathi, Babu, and Longvah (2018). Ghosh, Datta, and Datta (2019) reported a Vit B2 range 0.4- 1.4 μ g/g, and it is similar to this study, but the other vitamins are lower than the range. The vitamin B1 content in RRC is 0.55 mg/kg, and the vitamin B2 and vitamin B6 content in LK1-0646984 is 0.22 mg/kg and 4.06 mg/kg, which is slightly different from those studied by Abbas et al. (2011). Deepa, Singh, and Naidu (2008) reported the findings of medicinal brown rice from Njavara and non-medicinal rice varieties, which is closer to this study.



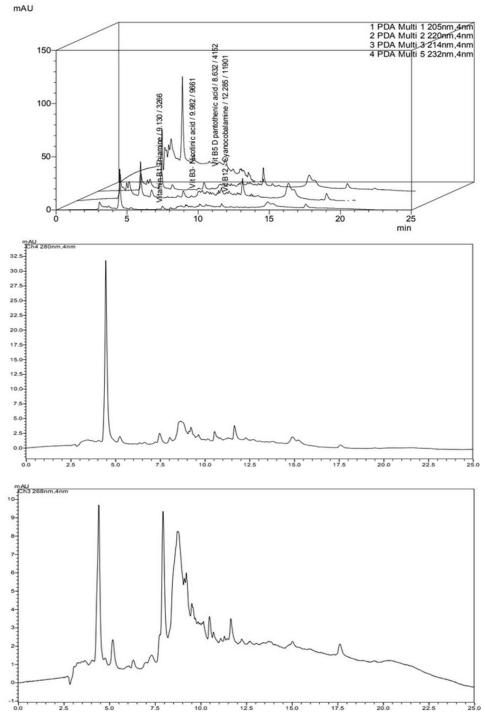
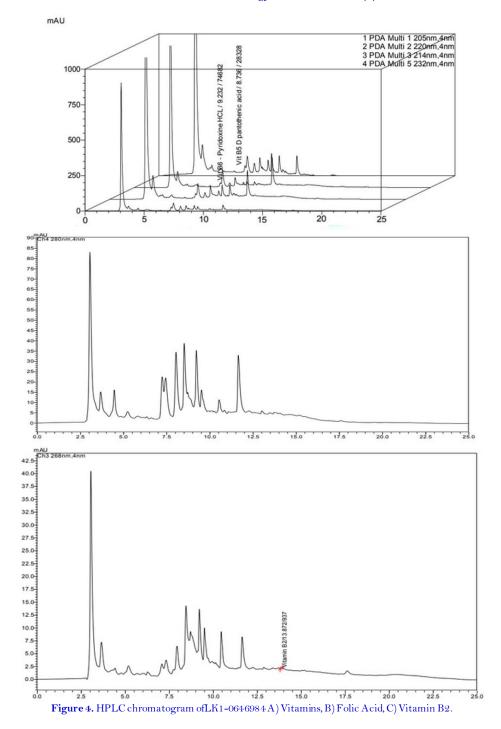


Figure 3. HPLC chromatogram of	f RC-0648663 A) Vitamin:	s, B) Folic Acid, C	C) Vitamin B2.
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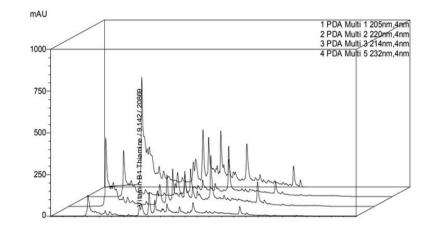
Sl. no.	Name	Ret. time (Min)	Area	Conc.(mg/Kg)
1	Vitamin B1	9.1	3,266	0.55
2	Vitamin B2	13.8	-	BDL
3	Vitamin B3	9.9	9,661	0.61
4	Vitamin B5	8.6	4,152	0.96
5	Vitamin B6	9.2	-	BDL
6	Vitamin B9	9.1	-	BDL
7	Vitamin B12	12.2	11,901	0.84
8	Vitamin E	8.2	-	BDL

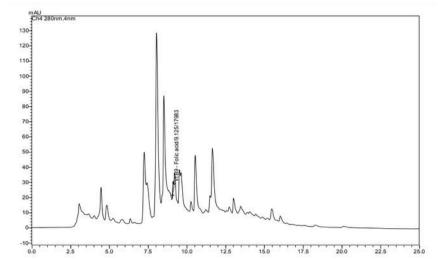


Sl. no.	Name	Ret.time(min)	Area	Conc.(mg/Kg)
1	Vitamin B1	9.1	-	BDL
2	Vitamin B2	13.8	937	0.22
3	Vitamin B3	9.9	-	BDL
4	Vitamin B5	8.7	28,328	6.52
5	Vitamin B6	9.2	74,682	4.06
6	Vitamin B9	9.1	-	BDL
7	Vitamin B12	12.2	-	BDL
8	Vitamin E	8.2	-	BDL

Note: BDL – Below detection level.

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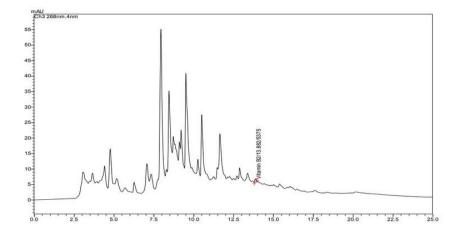


Figure 5. HPLC chromatogram of LK1-0646984HA) Vitamins, B) Folic Acid, C) Vitamin B2.

Sl.no.	Name	Ret.time(min)	Area	Conc.(mg/Kg)
1	Vitamin B1	9.1	20,869	3.53
2	Vitamin B2	13.8	5,375	1.26
3	Vitamin B3	9.9	-	BDL
4	Vitamin B5	8.7	-	BDL
5	Vitamin B6	9.2	-	BDL
6	Vitamin B9	9.1	17,983	1.16
7	Vitamin B12	12.2	-	BDL
8	Vitamin E	8.2	-	BDL

Table 5. Water soluble vitamins and Fat-soluble vitamins in LK1-0646984H.

5. CONCLUSION

Out of all the rice varieties from various districts, there were variations in the proximate composition, mineral levels, and vitamin content. Rice varieties revealed an ample quantity of nutrients. The experiment showed that LK1-0646984 has a higher nutritional value, more phytochemicals, and a higher antioxidant capacity. This may be because it helped seeds germinate. Considering the facts, it can be stated that both the rice varieties and husk as a supplement can be used for the development of value-added fermented food due to their high concentration of minerals, phytochemicals, and vitamins. Moreover, both germinated and non-germinated rice can be consumed as a diet in its original form without converting it into a food product.

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Institutional Review Board Statement: The Ethical Committee of the Central Institute of Technology Kokrajhar, Assam, India has granted approval for this study on 25 February 2022 (Ref. No. CITK/IEC/24). Transparency: The authors state that the manuscript is honest, truthful, and transparent, that no key aspects of the investigation have been omitted, and that any differences from the study as planned have been clarified. This study followed all writing ethics.

Competing Interests: The authors declare that they have no competing interests.

Authors' Contributions: Conceptualization, L.K., S.R. and D.C.B.; methodology, L.K.; supervision, S.R. All authors have read and agreed to the published version of the manuscript.

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